USSN: 09/920,000 Notice to File MP

Title: Use of Fluorescent Molecular Beacons in the Detection of Methylated Nucleic Acids

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## Marked-up Version of Amendments:

1.

against at least target sequence in the Myf-3 gene. Preferable target sequences of the Myf-3 gene are as follows:

5' GCG GCG ACT CCG ACG CGT CCA GCC CGC GCT CC 3' (SEQ ID NO:

1)
5' TTA TAC CGC AGG CGG GCG AGC CGC GGG CGC TCG CT 3' (SEQ ID NO: 2)

5' CCG AGA GCC CTG CGG GGC CCG CCC TCC TGC TGG CG 3' (SEQ ID NO: 3)

In another embodiment of the invention the labeled oligonucleoitde sequence is directed against at least a target sequence in the glutathione-S-transferase (pi) gene. preferable target sequences of the glutathione-S-transferase (pi) gene would be as follows:

5' CTC CAG CGA CGG CCT CGC GGC CTC CGA GCC TTA TAA G 3' (SEQ ID NO: 4)

5' GGG GAC GCG GGC CGC GCG TAC TCA CTG GTG GCG A 3' (SEQ ID NO: 5)

2.

Molecular Beacon A	5'-(6FAM) <u>CGA GGC</u> GGG CTG GAC GCG TCG GAG <u>GCC TCG</u> (DABCYL)-3' ( <u>SEQ ID NO: 6</u> )
Molecular Beacon B	5'-(6FAM) <u>CGA GGG</u> GGC TGG ACG CGT CGG AG <u>C</u> <u>CTC G (DABCYL)</u> -3' ( <u>SEQ ID NO: 7</u> )
Complementary strand A	5'-CTC CGA CGC GTC CAG CCC G-3' (SEQ ID NO: 8)
Complementary strand B	5'-CTC CGA CGC GTC CAG CCC-3' (SEQ ID NO: 9)
5M	5'-GC <sub>m</sub> G GC <sub>m</sub> G ACT CC <sub>m</sub> G AC <sub>m</sub> G C <sub>m</sub> GT CCA GCC C <sub>m</sub> GC <sub>m</sub> GCT CC-3' (SEQ ID NO: 10)
Non-5M	5'- GCG GCG ACT CCG ACG CGT CCA GCC CGC GCT CC-3' (SEQ ID NO: 1)

 $C_m = 5$  methylated cytosines

5M = preferred Myf-3 target sequence, 5M methylated, Non-5M unmethylated (see page 12, line 8)

The stem sequences are underlined

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3.

## **Methylated Oligonucleotides**

5-5M ID NO 11)	5'-GCG GCG ACT CCG ACG CGT CCA GCC CGC GCT CC-3' (SEQ
4-5M <u>ID NO 12)</u>	5'-GCG GCG ACT CCG ACG CGT CCA GCC CGC GCT CC-3' (SEQ
3-5M ID NO 13)	5'-GCG GCG ACT CCG ACG CGT CCA GCC CGC GCT CC-3' (SEQ
2-5M ID NO 14)	5'-GCG GCG ACT CCG ACG CGT CCA GCC CGC GCT CC-3' (SEQ
1-5M <u>ID NO 15)</u>	5'-GCG GCG ACT CCG ACG CGT CCA GCC CGC GCT CC-3' (SEQ
	M = methylated cytosine residues
	NB. Reaction profiles showin in Fig.2

Modifications and variations such as would be apparent to the skilled addressee are considered to fall within the scope of the present invention.